

DROUGHT TOLERANCE STUDIES THROUGH WSSI AND STOMATA IN UPLAND COTTON

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Abstract

Water stress susceptibility index (WSSI) and stomatal conductance were used to determine the stress tolerance of 10 upland cotton cultivars during 2009 at Sindh Agriculture University, Tandojam, Pakistan. The experiment was conducted in split plot design with irrigations as main plots and cultivars as sub-plots. Two irrigation treatments were used i.e. one has two irrigations (water stress) and other has eight irrigations (non-stress). Analysis of variance revealed significant genotypic differences about WSSI for all the traits. Non-significant interaction between irrigations and cultivars for seed cotton yield and boll weight exhibited varieties stability over irrigation regimes, whereas significant interactions between above parameters for plant height and bolls per plant suggested genotypic instability over irrigation treatments for these traits. Overall, cultivars mean performance for all the traits in stress conditions was poor as compared to non-stress conditions, nevertheless some cultivars exhibited nonsignificant mean differences in both irrigation regimes, thus showing higher stress tolerance. The WSSI values of seed cotton yield as displayed in biplot revealed that cultivars CRIS-477, CRIS-483 and CRIS-486 were found highly susceptible to water stress. Cultivars CRIS-476, CRIS-482, CRIS-487 and NIAB-78 were characterized as highly susceptible with minimum production even under optimum irrigation conditions. Cultivar CRIS-9 was moderately tolerant as produced low production. However, cultivars CRIS-485 and CRIS-484 were found highly stress tolerant because of minimum WSSI value and lower stomatal conductance. Negative correlations between water stress and WSSI for seed cotton yield and plant height revealed that any increase in the degree of stress caused a corresponding decrease in WSSI.

Introduction

Worldwide sustainability and advancement of cotton yield are the major challenges for meeting impending threats of increasing world population in the face of diminution of arable land, depletion of water resources and environmental stresses. However, drought is one of the major yield-limiting stresses (Boyer, 1982; Ullah *et al.*, 2008). Pakistan is experiencing water scarcity since last many years. Selection of cotton cultivars which survive and give better yields in water stress conditions have greater scope in Pakistan and the world over at large because of shortage of irrigation water resources. Breeding programmes for varietal improvement are being routinely undertaken in optimum conditions due to the fact that faster and greater improvement in productivity or other traits is rather easier to achieve by selecting under optimum conditions (Rossielle & Hamblin, 1981).

It is also true that drought tolerance is a complex trait with no certainty as to which adaptive mechanisms are related to higher yields and what selection criteria are to be used for screening cotton cultivars tolerant to water stress conditions (Feres 1987). However, there are evidences that variability among genotypes exists for stress tolerance. Such variability can simply be measured by placing several cultivars under both optimum and water deficit conditions (Fisher and Maurer 1978). Blum (1979) related two philosophies to breed for higher yields under drought conditions. One is selection for high yields accepting the hypothesis that if the yield of a particular genotype is increased in optimum conditions, it may increase in non-optimum conditions also. If this hypothesis is not true then the valid method would be selecting genotypes in stress conditions and those giving higher yields may be chosen (Medersi & Jafferis, 1973).

Comparisons of stomatal responses in cultivars with contrasting agronomic properties have been reported (Roak & Quisenberry, 1977) but no conclusive evidence has emerged on defined relations between stomatal

properties and yield. Cotton has a C₃ carbon metabolism; however, its photosynthetic potential is relatively high but reductions in photosynthetic rate of cotton under water-limited environment is documented (Pettigrew, 2004). This reduction may be attributed to stomatal (Cornic, 2000; Flexas *et al.*, 2004) and non-stomatal factors (Ennahli & Earl, 2005). While groping genotypic variation for photosynthetic capacity, Leidi *et al.*, (1993) found enormity of genotypic variation for photosynthetic rate at boll formation stage in cotton germplasm. Considerable intra-specific variation for stomatal conductance in *Gossypium hirsutum* under water deficit has also been reported. Thus they conclude that leaf photosynthesis and stomatal conductance are potential indicators for drought tolerance in cotton.

To attempt such studies, Fisher & Maurer (1978) proposed the drought susceptibility index (DSI) to express the decline in yield of a cultivar under drought conditions with respect to mean reduction of all the cultivars under test. Fisher & Wood (1979) reported a high positive correlation between DSI and potential yield in wheat. In cotton, a modification of this index was used by Cook (1989) to study the behavior of several upland cotton cultivars and found significant differences among genotypes. Lopez (1998) used the same index and found significant variation among cultivars and a positive correlation between the DSI and potential yield. Rajamani (1994) screened 20 cotton genotypes in rain-fed and non-stress (irrigated) conditions and reported significant variations among genotypes for yield and also noticed some genotypes with higher yields in stress conditions. Rajeswari (1995) screened 30 cotton genotypes under rainfed conditions for screening to drought tolerance and found that three genotypes high yield potential in water stress conditions. Ullah *et al.*, (2008) reported that seed cotton yield and biological yield were distinctly affected in all cultivars except few which proved their superiority to others in drought tolerance.

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Despite the fact that cotton is grown on less than 2.8% of the total cropped area of the world, yet the economy of several countries of Latin America, Asia, and Africa are substantially dependent on cotton production (Fortucci, 2002). Cotton crop requires sufficient amount of water for its normal growth. Due to severe shortage of good quality irrigation water, the crop experiences severe water insufficiency, hence reductions in crop productivity. This can be more acute due to excessive withdrawal of ground water and depleting irrigation water resources in the future (Ullah *et al.*, 2008). Keeping in view the situations, when sometimes, our cotton crop receives one/two irrigations only creating stress conditions, have forced cotton breeders to develop stress tolerant genotypes. Therefore, the present studies were carried out to determine the differences in yield and yield contributing traits of ten upland cotton genotypes under water stress and optimum irrigation conditions by adapting WSSI and stomatal conductance as measures of stress tolerance.

Materials and Methods

Plant material and experimental procedure: Ten newly developed cotton (*Gossypium hirsutum* L.) cultivars viz., CRIS-476, CRIS-477, CRIS-482, CRIS-483, CRIS-484, CRIS-485, CRIS-486, CRIS-487, CRIS-9 and NIAB-78 (susceptible standard cultivar) varying in pedigree, morphological characters, yield potential and yield contributing traits, were studied under two irrigation regimes during 2009 at Sindh Agriculture University, Tandojam, Pakistan. In regime first, only two irrigations

were applied and were considered as water stress conditions. In regime second, eight irrigations were applied and was considered as optimum conditions. The irrigations were applied through controlled Siphon method. Each irrigation of 308.8 mm/ha was considered as normal quantity of irrigation water under Pakistan's conditions. The experiment was conducted in split plot design with four replications, considering irrigations as main plots and cultivars as sub-plots, with plot size of 14×3.2 m². The treatment with optimum conditions received 1st irrigation after 35 days of planting and subsequent irrigations at normal intervals of 15 days. Whereas water stress treatment received only two irrigations, first after 60 days of sowing and the second after 60 days of first irrigation. The plant and row spacing were kept at 30 and 75 cm, respectively. All the cultural practices were done as per recommended package for cotton production. The crop was grown under uniform conditions to minimize environmental variations to the maximum possible extent.

Traits measurement and statistical analysis: The formula developed by Fisher & Maurer (1978) with little modification in terminology was used to determine the WSSI of each cultivar as under:

$$WSSI = [1 - (Y_s/Y_p)]/S$$

where

Y_s = Seed cotton yield or any character in water stress treatment.

Y_p = Seed cotton yield (potential yield) or any character in optimum irrigation treatment.

$$S = \text{Stress intensity} = 1 - \frac{\text{Mean } Y_s \text{ of all genotypes in stress treatment}}{\text{Mean } Y_p \text{ of all genotypes in optimum irrigated treatment}}$$

Data were recorded on four traits viz., plant height (cm), boll weight (g), bolls per plant and seed cotton yield (kg ha⁻¹). Analysis of variances was performed over irrigations and cultivars for WSSI according to Gomez & Gomez (1984). Correlation coefficients between the water stress treatment and the corresponding WSSI were worked out. This method of calculating the correlations is actually the modification of Fisher & Maurer (1978) that correlated optimum conditions with DSI (drought susceptibility index). Stomatal conductance (mmol m⁻²s⁻¹) was determined through Porometer-AP4 (Delta Devices, Cambridge, UK).

Results

Analysis of variance and WSSI analysis: Mean squares indicated significant differences among 10 cultivars tested for all the characters studied (Table 1). Effect of irrigation regimes was also significant for all the traits. The interactions between irrigation × cultivar seed cotton yield and boll weight were non-significant indicating that cultivars were fairly stable over irrigation regimes for these traits. However, irrigation × cultivar interaction for plant height, bolls per plant and stomatal conductance was significant indicating differential response of cultivars over irrigation treatments.

Table 1. Mean squares for yield and physiological traits in upland cotton cultivars.

Source of variation	d.f.	Mean squares				
		Stomatal conductance	Plant height	Bolls plant ⁻¹	Boll weight	Seed cotton yield
		4387346.7**	4712.45**	138.864**	1.058**	24982830.45**
Error (a)	6	552.07	6.650	0.338	0.050	397540.16
Cultivars (C)	9	103839.2**	312.117**	86.757**	0.825**	1116646.24**
I x C	9	29179.0**	87.450**	70.986**	0.032	148365.84
Error (b)	54	199.4	1.169	0.197	0.050	213171.89
Correlations (r) between water-stress condition and WSSI		-	r = -0.86**	r = -0.22	r = -0.20	r = -0.93**

** Significant at p 0.01

The WSSI values were calculated for determining the tolerance of genotypes under water stress (Table 2). The cultivars showing WSSI values less than 1.0 are more tolerant to water stresses while those with values above 1.0 are susceptible to stresses. In our situation, genotypes showing WSSI values less than 1.0 were CRIS-485 (0.432), CRIS-9 (0.787) and CRIS-484 (0.872), hence these genotypes were considered as more stress tolerant as

regards their yield. The smallest WSSI value (0.432) however was shown by CRIS-485, thus presenting the highest stress tolerance as compared to other cultivars. This was also supported by performance of cultivar CRIS-485 for plant height or boll weight with lowest WSSI values of 0.215 and 0.196, respectively. However, for boll weight, CRIS-485 (0.614) had 4th lowest value of WSSI in the series of ten cultivars.

Table 2. Water stress susceptibility index (WSSI) of seed cotton yield and its components.

Cultivars	Seed cotton yield	Bolls plant ⁻¹	Plant height	Boll weight
CRIS-476	1.235	1.414	0.905	1.429
CRIS-477	1.088	1.389	0.581	1.502
CRIS-482	1.233	0.397	1.028	0.837
CRIS-483	1.084	0.027	1.545	0.978
CRIS-484	0.872	0.972	0.966	1.451
CRIS-485	0.432	0.614	0.215	0.196
CRIS-486	1.338	1.073	1.641	0.275
CRIS-487	1.041	2.288	0.862	1.515
CRIS-9	0.787	0.482	1.323	1.263
NIAB-78	1.027	0.896	0.948	0.878

For reaching to the clear picture of stress tolerance, a biplot (Fig. 1) is also drawn and is divided into four quadrants. In biplot, quadrant-I represents those cultivars which are highly susceptible to stress but produced high production (above grand mean) in optimum irrigation conditions, and cultivars CRIS-477, CRIS-484 and CRIS-486 fall in this group. Quadrant-II corresponds to susceptible genotypes with lower yields, and cultivars

CRIS-476, CRIS-482, CRIS-487 and NIAB-78 represent this group. Quadrant-III includes genotypes which are fairly tolerant to water stress but produced lower production thus CRIS-9 fits in this group. Quadrant-IV demonstrates those genotypes which are not only highly stress tolerant but simultaneously give maximum production. The cultivars CRIS-484 and CRIS-485 come in this group.

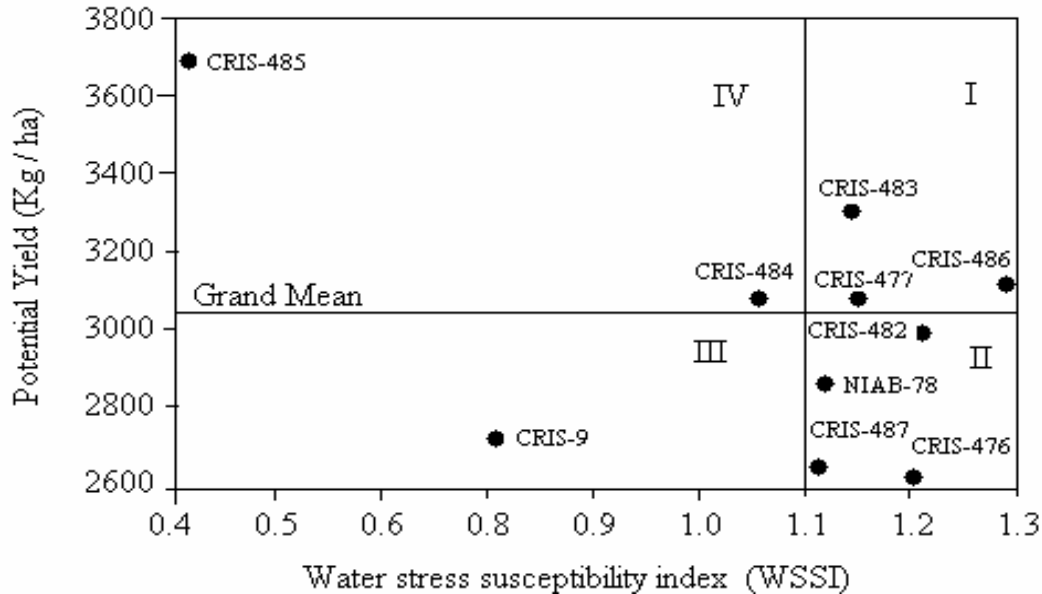


Fig. 1. Biplot between maximum production under optimum irrigation conditions and WSSI values for upland cotton genotypes.

Stomatal conductance and mean performance under both irrigation regimes: The stomatal conductance in non-stress conditions ranged from 450 to 870, while it varied from 135 to 520 $\text{mmol m}^{-2}\text{s}^{-1}$ in water stress conditions (Table 3). Cultivar CRIS-485 with minimum stomatal conductance ($135 \text{ mmol m}^{-2}\text{s}^{-1}$) gave maximum yield (3133.5 kg/ha) because it had less transpiration rate per unit area and per unit time. It was followed by cultivar CRIS-484 with yield of $2067.5 \text{ kg ha}^{-1}$ and stomatal

conductance was $250 \text{ mmol m}^{-2}\text{s}^{-1}$. Generally cultivars with higher stomatal conductance in stress conditions yielded relatively lower. Results indicated that cultivars which gave higher yields in stress conditions showed lower stomatal conductance and thus were more drought tolerant (Table 3). In other words, there was a negative association ($r = -0.93$) between cultivars drought tolerance and stomatal conductance (Table 1).

Table 3. Mean performance of upland cotton cultivars for seed cotton yield and its components in non-stress and water-stress conditions.

Cultivars	Stomatal conductance (mmol m ⁻² s ⁻¹)		Plant height (cm)		Bolls plant ⁻¹ (#)		Boll weight (g)		Seed cotton yield (kg ha ⁻¹)	
	NS	WS	NS	WS	NS	WS	NS	WS	NS	WS
CRIS-476	870	520	110.5	97.5	38.4	30.0	3.2	2.9	2689.5	1463.3
CRIS-477	770	420	116.5	107.8	35.3	27.7	3.3	2.9	3029.0	1811.8
CRIS-482	780	430	123.5	107.0	31.1	29.2	2.9	2.7	2994.5	1632.3
CRIS-483	680	330	114.5	91.5	23.8	23.9	3.6	3.4	3312.3	1887.0
CRIS-484	600	250	117.5	102.8	29.8	25.4	3.2	2.8	3040.3	2067.5
CRIS-485	450	135	125.0	121.5	33.5	30.4	3.7	3.6	3727.5	3133.5
CRIS-486	860	500	119.5	94.0	38.3	31.9	2.6	2.5	3048.0	1542.8
CRIS-487	780	435	120.5	107.0	35.7	23.2	3.3	2.9	2766.3	1703.0
CRIS-9	790	437	123.5	102.3	32.1	29.7	3.3	3.0	2779.0	1971.8
NIAB-78*	750	498	111.5	97.8	35.5	30.6	2.9	2.7	2887.5	1792.5
Average	733	396	118.3	102.9	33.4	28.2	3.2	2.9	3027.4	1900.6
% RR in WS	-	46.0	-	13.02	-	17.36	-	6.25	-	37.22
LSD (5%) Treatments	12.9		1.41		0.32		0.12		345.0	
LSD (5%) Cultivars	14.1		1.08		0.44		0.22		461.0	
LSD (5%) Treatments x Cultivars	20.0		1.52		0.62		N.S.		N.S.	

% RR = Average percentage relative reduction in water-stress conditions

NS = Non-stress, WS = Water-stress, N.S. = Non-significant

* = Known water stress susceptible cultivar

Results in Table 3 also depicted the mean performance of cultivars in water stress and non-stress conditions. On average, the cultivars performed poorly in stress treatment as compared to non-stress treatment for all the traits. Overall, plant height of all the cultivars suffered in stress conditions which ranged from 91.5 to 121.5 cm as compared to increased height (110.5 to 125.0 cm) in non-stress conditions (Table 3). Plant height as an average of ten cultivars was 102.9 cm in stress conditions as compared to 118.3 cm in non-stress conditions. Cultivars CRIS-476, CRIS-477, CRIS-484, CRIS-485, CRIS-487 and NIAB-78 which had less than 1.0 WSSI values sustained less water stress, yet minimum value was given by CRIS-485 being highly tolerant to water stress. However, the cultivar CRIS-485 maintained its plant height in both stress (121.5 cm) and non-stress (125.0 cm) conditions and that could be a probable reason of producing maximum yield.

All the cultivars also manifested lowest bolls per plant (23.2 to 31.9) in stress condition as compared to increased bolls per plant (23.8 to 38.4) in optimum irrigated conditions (Table 3). It holds true that average bolls per plant reduced to 28.2 in stress conditions against 33.4 bolls per plant in non-stressed conditions. The WSSI values suggested that six among ten cultivars manifested the values less than 1.0 suggesting their stress tolerance, yet the lowest WSSI values was expressed by cultivars CRIS-483 (0.027), CRIS-482 (0.397), CRIS-9 (0.482) and CRIS-485 (0.614).

Boll weight is regarded as an important yield component in cotton. All the cultivars showed small bolls except CRIS-485 in stress conditions (2.5 to 3.6 g) as compared to increased boll weight (2.6 to 3.7 g) in non-stress conditions (Table 3). Average boll weight, in stress conditions weighed 2.9 g as compared to 3.2 g in optimum irrigation conditions, which suggests that water deficit reduced the boll weight considerably. CRIS-485 being a highly tolerant cultivar to stress conditions also exhibited maximum boll weight in both stress (3.6 g) and non-stressed (3.7 g) irrigation conditions. Cultivars CRIS-482 (0.837), CRIS-483 (0.978), CRIS-485 (0.196), CRIS-486 (0.275) and NIAB-78 (0.878) gave WSSI values less

than 1.0 and were less susceptible to stress conditions (Table 2). However, smallest WSSI value of 0.196 was given by cultivar CRIS-485 being highly tolerant, whereas largest WSSI value of 1.515 recorded by CRIS-487 and was highly susceptible to water stresses.

In stress conditions, the seed cotton yield varied from 1463.3 to 3133.5 kg ha⁻¹ where maximum yield of 3133.5 kg ha⁻¹ was obtained from cultivar CRIS-485. However, in non-stress treatment, the yield ranged from 2689.5 to 3727.5 kg ha⁻¹ where the same cultivar also produced maximum yield of 3727.5 kg ha⁻¹, hence showing low water stress susceptibility as compared to other cultivars. On average, seed cotton yield of all the cultivars in stress conditions (1900.6 kg ha⁻¹) was too much low as compared to increase yield (3027.4 kg ha⁻¹) in optimum irrigation conditions, which suggests that water deficit eventually reduced the seed cotton yield. Cultivar CRIS-476 by having maximum WSSI value (1.235) was found more susceptible to water stress conditions which also gave lowest yield among the series of cultivars in both irrigations regimes.

Discussion

In both stress and non-stress conditions, the cultivar CRIS-485 showed low water stress susceptibility as compared to other cultivars. This cultivar has exhibited high stress tolerance because of small yield differences under both environments. Cultivar CRIS-476 was found more susceptible to water stress conditions due to WSSI value and which also gave lowest yield among all the cultivars in both irrigations regimes. Rajamani (1994) and Rajeswari (1995) observed some genotypes having high yield potential in water stress conditions while evaluating the cotton genotypes under rain-fed conditions. Ullah *et al.*, (2008) noticed that seed cotton yield and biological yield were distinctly affected in all cultivars, except few which proved their superiority over others in drought tolerance. Cultivars showing WSSI values less than 1.0 are more tolerant to water stresses while those with values above 1.0 are susceptible to stresses. In our situation,

genotypes showing WSSI values less than 1.0 were CRIS-485, CRIS-9 and CRIS-484, hence these genotypes were considered as more stress tolerant as regards their yield. The smallest WSSI value however was shown by CRIS-485, thus presenting the highest stress tolerance as compared to other cultivars in the test. Fisher & Maurer (1978) proposed the DSI to express the decline in yield of a cultivar under drought conditions, while Fisher & Wood (1979) also reported a high positive correlation between DSI and the potential yield.

According to biplot Quadrant-IV demonstrates that genotypes i.e., CRIS-484 and CRIS-485 not only highly stress tolerant but simultaneously provided maximum production. Rajamani (1994) screened 20 genotypes in rainfed and irrigated conditions and noticed significant differences among the genotypes for yield. He reported that two genotypes TKH680 and TKH679 produced higher yields in stress conditions with tolerance indices of 1.37 and 1.05, respectively. Rajeswari (1995) evaluated 30 genotypes under rainfed conditions for drought tolerance and found three genotypes with high yield potential and drought tolerance.

Results suggested that all the traits i.e. seed cotton yield, boll weight, plant height and bolls per plant exhibited negative correlations with WSSI whereas stomatal conductance had negative correlation with seed cotton yield only, suggesting that a unit increase in particular character in stress conditions caused a corresponding decrease in susceptibility index and stomatal conductance, thus resulting in increased stress tolerance. Nevertheless, only two of the four correlations i.e., seed cotton yield with WSSI and plant height with WSSI were significant indicating greater importance of yield and plant height traits in water stress tolerance studies. Gutierrez *et al.*, (1998) evaluated 25 upland cotton genotypes under drought and optimum irrigation conditions. They found that cultivars with high yields in optimum conditions, and drought tolerance were the ones with WSSI values less than 1.0. They used optimum conditions to calculate correlations of yield with its corresponding drought tolerance index, hence they recorded positive correlations and whereas we obtained negative correlations with similar interpretations. However, the method used in our studies is more justifiable and valid than their results.

Cultivars (CRIS-485 and CRIS-484) with minimum stomatal conductance gave maximum yield because of less transpiration per unit area and time. Generally cultivars with higher stomatal conductance in stress conditions yielded relatively lower. Results revealed that due to negative association between cultivars drought tolerance and stomatal conductance, the cultivars manifested higher yields in stress conditions with lower stomatal conductance and thus were more drought tolerant. Therefore, stomatal conductance and leaf photosynthesis could be potential indicators for drought tolerance in cotton. Pettigrew (2004) documented the reduction of photosynthetic rate in cotton under water-limited environment. Cornic (2000) and Flexas *et al.*, (2004) were of the opinion that this reduction may be attributed to stomatal, and non-stomatal factors (Ennahli & Earl, 2005). Leidi *et al.*, (1993) also found enormity of genotypic variation for photosynthetic rate at boll formation stage and considerable intra-specific variation

for stomatal conductance in *Gossypium hirsutum* L., germplasm. On the contrary, Roak & Quisenberry (1977) were of the view that although assessment of stomatal responses in cultivars with contrasting agronomic properties have been reported but no conclusive evidence has emerged on defined relations between stomatal properties and yield. Such contradictions may be due to different plant material used under distinct climatic conditions.

Plant height also determines the yield in the sense that as plant height increases, both the number of fruiting branches and fruiting points also increases, consequently yield also increases. Cultivar CRIS-485 maintained its plant height in both stress and non-stress conditions that could be a probable reason of its producing maximum yield. In water stress conditions, plant height decreased significantly as compared to optimum irrigation conditions (CCRI, 2009). CRIS-485 being a highly tolerant cultivar to stress conditions also exhibited maximum boll weight in both stress and non-stressed conditions. According to WSSI values, cultivar CRIS-485 being highly tolerant has smallest values as compared to CRIS-487 which was highly susceptible to water stresses with largest WSSI value. Water stress conditions decreased the boll weight as compared to optimum irrigation conditions (CCRI, 2009). Maurer (1991) also evaluated two cultivars and found that boll weight reduced due to stress conditions. Bolls per plant is also directly related to seed cotton yield and has a major role in managing seed cotton yield. It was assumed that in water stress conditions, the cultivars shed flowers and set small bolls, consequently the yield losses occurred. The WSSI values suggested that six cultivars (CRIS-483, CRIS-482, CRIS-9 and CRIS-485) manifested the values less than 1.0 suggesting their stress tolerance. Bolls per plant and eventually seed cotton yield were badly affected in water stress as compared to optimum irrigation conditions (CCRI, 2009). Maurer (1991) also noticed reduced bolls per plant in stress conditions.

Conclusion

Cultivars CRIS-485 and CRIS-484 were more water stress tolerant as compared to other cultivars by displaying less than 1.0 WSSI values for most of the characters studied. Hence, these two cultivars could either be used in the areas which experience shortage of water or in hybridization programmes to develop new water stress tolerant genotypes.

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